

IONIC STRENGTH CONTROL OF THE SPATIAL DISTRIBUTION OF BACTERIAL CELLS DURING AN *IN SITU* RESTING-STATE BIOAUGMENTATION EXPERIMENT

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The widespread use of chlorinated solvents and the subsequent disposal of spent solvents to the environment has resulted in contaminated groundwater at many government and industrial facilities. Because standard remediation methods can be costly and frequently ineffective, a novel resting-state bioaugmentation technique was developed and field tested as an alternative. This technique employs a naturally-occurring species of methanotrophic bacteria (*Methylosinus trichosporium* OB3b). Bacteria, grown in surface bioreactors, are injected into the contaminated aquifer to establish an *in situ* fixed-bed bioreactor. In the field test, a biofilter was generated at a depth of 27 m by injecting 5.4 kg (dry weight) of the bacteria suspended in 1800 L of uncontaminated groundwater (5.4×10^9 cell/mL). Approximately 50% of the injected bacteria attached to the aquifer material. Groundwater was subsequently extracted from the well, drawing contaminated groundwater through the biofilter region. Measurements confirmed substantial bioremediation, with the background contaminant level (425 ppb TCE) reduced by as much as 99% during the early stages of the test. Results from earlier tracer tests indicated a highly heterogeneous subsurface environment that presented a difficult challenge for the *in situ* biodegradation application and provided a basis for determining the extent of biodegradation achieved during the test. In addition to the heterogeneities determined from the tracer data, the chemical environment varied as a function of distance from the injection well due to the sorption and precipitation of the pH buffer added to the bacterial suspension — substantially reducing the ionic strength of the solution. Because bacterial attachment efficiency is a strong function of ionic strength, the resulting changes were inferred to influence the distribution of bacteria in the subsurface. These results help interpret the field data and may provide a mechanism to engineer the spatial distribution of bacteria in the subsurface by controlling the ionic strength of the injected fluid phase.

Keywords: bioremediation, bioaugmentation, methanotroph, in-situ, solvents

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